

Claims

WHAT IS CLAIMED IS:

- 5 1. A method for producing a transgenic Gramineae plant comprising the steps of:
- 10 (a) isolating a zygote from a Gramineae plant to be transformed in a way that said isolated zygote becomes substantially free from its naturally surrounding tissue,
- (b) introducing a DNA composition comprising a genetic component into the genome of said Gramineae plant, wherein said introduction is mediated by Agrobacterium transformation into said isolated zygote;
- 15 (c) regenerating Gramineae plants from said zygotes which have received said genetic component; and
- (d) identifying a fertile, transgenic Gramineae plant whose genome has been altered through the stable introduction of said genetic component.
- 20 2. The method of claim 1, wherein the Gramineae plant is selected from the group consisting of wheat, maize and barley.
3. The method of claim 1 or 2, where in the Gramineae plant is a Triticum species.
- 25 4. The method of any of claim 1 to 3, wherein the Gramineae plant is regenerated from said isolated zygote by a method comprising co-cultivation of said isolated zygote and/or the zygotic embryo derived therefrom with a feeder system.
- 30 5. The method of any of claim 1 to 4, wherein the Gramineae plant is regenerated from said isolated zygote by a method comprising co-cultivation of said isolated zygote and/or the zygotic embryo derived therefrom with a culture of isolated immature pollen or pistils.
- 35 6. The method of any of claim 1 to 5, wherein the Gramineae plant is regenerated from said isolated zygote by a method comprising co-cultivation of said isolated zygote and/or the zygotic embryo derived therefrom with
- 40 a) a culture of androgenetically developing barley pollen or
b) a culture of wheat or barley pistils or
c) any combination of a) and b).
7. The method of any of claim 1 to 6, wherein the zygotes and the feeder system are physically separated in a way to prevent mixing of the different cell types but to allow exchange of growth factors, proteins, media components, and other low molecular weight compounds.
- 45 8. The method of any of claim 1 to 7, wherein co-cultivation of the zygotes and the feeder system are employed already during Agrobacterium co-cultivation in a way

that the co-cultivation culture of the zygotes and Agrobacterium is physically separated from the feeder system in a way to prevent contact of the Agrobacteria with the feeder system but to allow exchange of growth factors, proteins, media components, and other low molecular weight compounds.

5

9. The method of any of claim 1 to 8, wherein said genetic component is transmitted through a complete sexual cycle of said transgenic Gramineae plant to its progeny, wherein said progeny does not comprise a selectable or screenable marker gene.

10

10. The method of any of claim 1 to 9, wherein said method does not comprise a step which leads to dedifferentiation of the zygote or the zygote-derived embryo.

15

11. The method of any of claim 1 to 10, wherein said genetic component comprises a expression cassette comprising a nucleic acid sequence operably linked to a promoter active in said Gramineae plant, wherein expression of said nucleic acid sequence confers a phenotypically distinguishable trait to said Gramineae plant.

20

12. The method of any of claim 1 to 11, wherein the pH of the medium used during co-cultivation of the isolated zygote with Agrobacterium is kept in a range from about 5.8 to about 6.0.